Proposal Reviews

#214: Decreased Survival of Chinook Salmon Smolt in the Old River: Biological Responses to Contaminants

University of California, Davis, Bodega Marine Laboratory

Research and Restoration Technical Panel Review	
Delta Regional Review	
External Scientific Review	#1 #2 #3 #4
Environmental Compliance	
Budget	

Research and Restoration Technical Panel Review:

CALFED Bay-Delta 2002 ERP PSP
Research and Restoration Technical Panel Review Form

Proposal Number: 214

Applicant Organization: University of California, Davis, Bodega Marine Laboratory

Proposal Title: Decreased Survival of Chinook Salmon Smolt in the Old River: Biological Responses to Contaminants

Review:

Please provide an overall evaluation summary rating:

Superior: outstanding in all respects;
Above Average: Quality proposal, medium or high regional value, and no significant administrative concerns;
Adequate: No serious deficiencies, no significant regional impediments, and no significant administrative concerns;
Not Recommended: Serious deficiencies, significant regional impediments or significant administrative concerns.

Overall Evaluation Summary Rating	**Provide a brief explanation of your summary rating**
-Superior	**The experimental approach was considered to be inappropriate. Moreover, this is a high risk proposal due to the variable nature of river management.**
-Above average	
-Adequate	
XNot recommended	

1. **Goals and Justification.** Does the proposal present a clear statement of goals, objectives and hypotheses? Does the proposal present a clear justification and conceptual model for the project?

   The overall objective in this proposal is to determine whether toxicants could play a role in the decreased survival of chinook salmon smolts that pass down the Old River compared with those that pass through the San Joaquin River during their migration to the Pacific Ocean. Currently, contaminants have not been measured in the Old River during the VAMP period despite the effort that has gone in to constructing the barrier at the head of the Old River in an attempt to by pass the Old River during a period of time that it is heavily influenced by agriculture pesticides. Also, the authors collected data for this study in 2001 that was to be supported by CALFED at that time but was not funded due to an ongoing dispute between CALFED and the University of California.

The justification is strong for measuring contaminant loads in fish, but the measurement of the biomarkers is far less justified, due to the non-specific nature of many of the responses to be quantified, as noted in the external reviews.

2. **Likelihood of Success (Approach, Feasibility, Capabilities and Performance Measures).** Is the project likely to succeed based on the approach, feasibility and project team capabilities? Are the proposed performance measures adequate for measuring the project's success?

   The panel felt that the approach for measuring contaminant loads (i.e. caging studies) was not appropriate, due to many of the problems (including high mortalities) that the investigators previously experienced. It seems that a much needed precursor to any of this work would be assessment of contaminant exposure in resident fish species, to avoid the caging issues. In any event, caging the fish for only 4 days may not be adequate for certain responses to be manifest. This is a capable research team, and performance measures are appropriate.

3. **Outcomes and Products.** Will the project advance the state of scientific knowledge in general and/or make an important contribution to the state of knowledge of the Bay-Delta Watershed? For restoration proposals, is the project likely to contribute to ecosystem restoration or species recoveries in a significant way? Will the project produce products useful to decision-makers and scientists?

   This study would not be likely to provide useful data as to the potential extent of contaminant exposure of outmigrating juvenile salmon in this highly modified and impacted system. The biological endpoints are not likely to be clear.

4. **Cost/Benefit Comments.** Is the budget reasonable and adequate for the work proposed?

   The budget seems reasonable for an 18 month study.

5. **Regional Review.** How did the regional panel(s) rank the proposal (High, Medium, Low)? Did the regional panel(s) identify significant benefits (regional priorities, linkages with other activities, local involvement) or impediments (local constraints, conflicts with other activities, lack of local involvement) to this proposal? What were they?

   This proposal was ranked low by the one region which provided a review. It was pointed out that the management of the river during the VAMP, especially the timing of the HORB construction, may preclude this study from being conducted.

6. **Administrative Review.** Were there significant concerns about the proposal with regard to the prior performance, environmental compliance and budget administrative reviews? What were they?

   No significant issues raised

Miscellaneous comments:

None

Delta Regional Review:

Proposal Number: 214

Proposal Title: Decreased Survival of Chinook Salmon Smolt in the Old River: Biological Responses to Contaminants

Overall Ranking: **X**Low **-**Medium **-**High

Provide a brief summary explanation of the committee's ranking:

Exploring role of toxicants in decreased survival of chinook salmon smolts passing through Old River isn't a priority when compared to other documented causes of mortality.

1. Is the project feasible based on local constraints?

   XYes **-**No

   How?

   Feasibility studies have been conducted so the mechanics of the test itself is possible. However, there are conditions that are very possible that are beyond the control of the project proponents such as: a. Delays in the construction of the HORB has occurred often in the past. Without the installation of the HOR Barrier, the project could not be completed. b. It is not uncommon for extreme weather conditions (unseasonal high temperatures especially) that affect fish mortality to occur.

2. Does the project pursue the restoration priorities applicable to the region as outlined in the PSP?

   XYes **-**No

   How?

   Restore shallow water habitats in the Delta for the benefit of at-risk species while minimizing potential adverse effects of contaminants. -effect of contaminants - fish survival in Central and South Delta However, the relative significance of toxics effects relative to other concerns in the South Delta are unclear.

3. Is the project adequately linked with other restoration activities in the region, such as ongoing implementation projects and regional planning efforts?

   -Yes **X**No

   How?

   Evaluates the effectiveness of VAMP in the context of contaminants but needs to strengthen discussions with those working with resident fish and hydrodynamics in the south Delta to broaden application of work.

4. Does the project adequately involve local people and institutions?

   -Yes **X**No

   How?

   Project is a multi agency collaboration on field (USFWS), lab (USGS) and data analysis(Bodega Marine Lab) but should broaden discussions of possible applications and implications of work with others doing work in the area.

Other Comments:

Should have a stronger hydrodynamics component evaluating water residence time, tidal effects, water circulation patterns.

External Scientific: #1

Research and Restoration External Scientific Review Form

Proposal Number: **214**

Applicant Organization: **University of California, Davis, Bodega Marine Laboratory**

Proposal Title: **Decreased Survival of Chinook Salmon Smolt in the Old River: Biological Responses to Contaminants**

Conflict of Interest Statements:
I have no financial interest in this proposal.
XCorrect
-Incorrect

In the blank below please explain any connection to proposal, to applicant, co-applicant or subcontractor or to submitting institution (write "none" if no connection):

none

Review:

Please provide an overall evaluation summary rating:

<u>Excellent:</u> outstanding in all respects;
<u>Good:</u> quality but some deficiencies;
<u>Poor:</u> serious deficiencies.

Overall Evaluation Summary Rating	**Provide a brief explanation of your summary rating**
-Excellent	**Good-The theme has merit for addressing CALFED goals. There is a possibility of results that will be difficult to interpret and apply in a management sense.**
XGood	
-Poor	

1. **<u>Goals.</u>** Are the goals, objectives and hypotheses clearly stated and internally consistent? Is the concept timely and important?

   The goals are clearly stated in the problem statement, namely to determine the role of contaminants in the decreased survival of Chinook salmon smolts that pass down the Old River verses those in the San Joaquin.

2. **<u>Justification.</u>** Is the study justified relative to existing knowledge? Is a conceptual model clearly stated in the proposal and does it explain the underlying basis for the proposed work? Is the selection of research, pilot or demonstration project, or a full-scale implementation project justified?

A conceptual model is outlined in the text and as a figure (3). The figure doesnt do service as a conceptual model and the overall conceptual model focuses just on contaminants as a factor in the survival of Chinook salmon in Old river. This is a research project that is focused on comparative field experiments based upon reported differences in escapement between two of the major river systems. It would have been nice if the proposal stated how much these differences have been estimated to be and what kinds of effects the VAMP project have had on the recruitment process for salmon.

3. **Approach.** Is the approach well designed and appropriate for meeting the objectives of the project? Are results likely to add to the base of knowledge? Is the project likely to generate novel information, methodology or approaches? Will the information ultimately be useful to decision-makers?

   I believe that the study of contaminants is warranted in this system and that the basic analytical methods are well suited to such a study. I have some concerns about the other aspects of the approach, specifically about the caging studies. Caging studies are often difficult to execute without confounding or confusing results. I understand the appeal in terms of the difficulty of knowing exactly what the suite of chemical will be in advance. Also a comparative approach may have some merit if the caging is done properly. The weakness is that you just cant ignore the fact that a caged fish does not behave the same as a free roaming individual and you also never know exactly what the caging effect has been. Another problem is that the fish are not feeding as they would and not migrating or responding to their environment in a natural way. The danger is that a cage study can provide some pitfalls for a poorly executed field exposure experiment. Chances are you wont know it until its too late to reproduce. My other concern is that the proposal begins by justifying this research because of the differences in survival of salmon between the Old River and the San Joaquin. The preliminary caging study doesnt seem to show that difference (figure 5), but there were significant mortalities apparently associated with caging or temperature. This study also doesnt consider that the major effect may be on the prey of the salmon and not a direct effect. I noticed that colleagues are already looking at the prey issue in a contaminant study that is already funded. There have got to be some alternative theories to the contaminant effects as the cause of poor survival in the Old River. It would have been nice to see those discussed in this proposal and an argument made in that context for further studies of contaminant effects with salmon.

4. **Feasibility.** Is the approach fully documented and technically feasible? What is the likelihood of success? Is the scale of the project consistent with the objectives?

   Sublethal effects on organism may be the most important ones because they go unrealized until we see major declines in populations. They are also the most difficult to assess. The technical feasibility of the study is high but the probability of clearly determining the population level effects of contaminants on salmon smolts, even in a relative sense, is questionable.

5. **Project-Specific Performance Measures.** Does the project include appropriate performance measures to measure success relative to the project's goals and objectives? Is there enough detail as to how the performance measures will be quantified? For restoration projects, are monitoring plans explicit and detailed enough to determine if performance measures will be adequately assessed?

   The usual performance measures section is listed.

6. **Products.** Are products of value likely from the project? Specifically for restoration projects, are products of value also likely from the monitoring component? Are interpretative outcomes likely from the project?

Several of the typical products are listed. Results from this project could be valuable to CALFED, but interpretive outcomes could prove elusive in terms of management standards for contaminants.

7. **Capabilities.** What is the track record of applicants in terms of past projects? Is the project team qualified to efficiently and effectively implement the proposed project? Do they have available the infrastructure and other aspects of support necessary to accomplish the project?

   The team has the background to conduct the proposed study and the infrastructure is available for the analyses.

8. **Cost/Benefit Comments.** Is the budget reasonable and adequate for the work proposed?

   The budget is probably reasonable for the amount of activities in terms of laboratory analyses. The field work is only slated for a single year. With only one year of field sampling it will be difficult to interpret outcomes, especially if it happens to be a particularly good year or bad year with respect to river flow.

Miscellaneous comments:

External Scientific: #2

Research and Restoration External Scientific Review Form

Proposal Number: **214**

Applicant Organization: **University of California, Davis, Bodega Marine Laboratory**

Proposal Title: **Decreased Survival of Chinook Salmon Smolt in the Old River: Biological Responses to Contaminants**

Conflict of Interest Statements:
I have no financial interest in this proposal.
XCorrect
-Incorrect

In the blank below please explain any connection to proposal, to applicant, co-applicant or subcontractor or to submitting institution (write "none" if no connection):

none

Review:

Please provide an overall evaluation summary rating:

Excellent: outstanding in all respects;
Good: quality but some deficiencies;
Poor: serious deficiencies.

Overall Evaluation Summary Rating	**Provide a brief explanation of your summary rating**
XExcellent	**This is important research, the proposal is well put together. There are some experimental design considerations that the investigators should consider.**
-Good	
-Poor	

1. **Goals.** Are the goals, objectives and hypotheses clearly stated and internally consistent? Is the concept timely and important?

   Rating: Very good The overall objective in this proposal is to determine whether toxicants could play a role in the decreased survival of chinook salmon smolts that pass down the Old River compared with those that pass through the San Joaquin River during their migration to the pacific ocean. Currently, contaminants have not been measured in the Old River during the VAMP period despite the effort that has gone in to constructing the barrier at the head of the Old River in an attempt to by pass the Old River during a period of time that it is heavily influenced by agriculture pesticides. Also, the authors collected data for this study in 2001 that was to be supported by CALFED at that time but was not funded due to an ongoing dispute between CALFED and the University of California.

2. **Justification.** Is the study justified relative to existing knowledge? Is a conceptual model clearly stated in the proposal and does it explain the underlying basis for the proposed work? Is the selection of research, pilot or demonstration project, or a full-scale implementation project justified?

   Rating: Very good Yes, the study is justified relative to existing knowledge. The use of biomarkers to examine sublethal effects of salmon smolts due to contaminants have been used successfully in salmon smolts from Puget Sound and was referred to by the applicants. These studies will provide information to CALFED on an economically important endangered species and at what level contaminants may influence salmon populations. This research is an important prelude to any habitat restoration efforts.

3. **Approach.** Is the approach well designed and appropriate for meeting the objectives of the project? Are results likely to add to the base of knowledge? Is the project likely to generate novel information, methodology or approaches? Will the information ultimately be useful to decision-makers?

   Rating: Very good. The authors do a very good job in explaining why they think there may or may not be differences in sublethal effects during the VAMP, pre- and post-VAMP periods. This reviewer would be interested in knowing if indeed it is possible to determine that survival of fish going through the Old River during the VAMP period is actually lower than those going through the Old River during Pre and Post VAMP periods where contaminant exposure may be less. Is data on adult returns actually detailed enough to determine this information? Also, at one point the authors say that at post VAMP the Old River is dominated by Sacramento water quality that they predict to be less harmful than the agriculture return flows which dominate the San Joaquin River. However, in the paragraph that follows this assertion, it is stated that during the VAMP period the Sacramento River contains pesticides from rice fields. Is this not the case during the post VAMP period? Also during the VAMP period while the old river is being influenced by these pesticides, San Joaquin is still receiving molinate, thiobencarb, and carbofuran.

   Although new techniques are not being developed, the information that will be generated is important to decision-makers.

4. **Feasibility.** Is the approach fully documented and technically feasible? What is the likelihood of success? Is the scale of the project consistent with the objectives?

   Rating: Very Good The approach is very feasible and the objectives are within the grasp of these competent researchers. However, the authors may want to consider using more than 10 fish per net pen. This sample size seems very small and may affect their likelihood of success. Also, tissue samples for contaminant analysis should be taken, as well as taking water samples throughout the experiment. Additionally they may want to consider sampling fish throughout the 3 critical periods instead of one sampling after 4 days of exposure during each period. It seems odd that the authors dont need transport permits or special permits to allow them to put holding cages and nonresident fish in to the two rivers.

5. **Project-Specific Performance Measures.** Does the project include appropriate performance measures to measure success relative to the project's goals and objectives? Is there enough detail as to how the performance measures will be quantified? For restoration projects, are monitoring plans explicit and detailed enough to determine if performance measures will be adequately assessed?

Rating: Very good

6. **Products.** Are products of value likely from the project? Specifically for restoration projects, are products of value also likely from the monitoring component? Are interpretative outcomes likely from the project?

   Rating: Excellent Valuable products are the likely outcome of this research. Seven products are expected from this work in the form of presentations, reports and publications. Although contributions to larger data management systems has not been addressed the authors do intend to work cooperatively with state and federal agencies.

7. **Capabilities.** What is the track record of applicants in terms of past projects? Is the project team qualified to efficiently and effectively implement the proposed project? Do they have available the infrastructure and other aspects of support necessary to accomplish the project?

   Rating: Excellent Dr. Anderson and her team have an excellent track record and reputation for their expertise in toxicology.

8. **Cost/Benefit Comments.** Is the budget reasonable and adequate for the work proposed?

   Rating: Excellent

   The budget is very reasonable and adequate for an eighteen-month study.

Miscellaneous comments:

External Scientific: #3

Research and Restoration External Scientific Review Form

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Conflict of Interest Statements:
I have no financial interest in this proposal.
XCorrect
-Incorrect

In the blank below please explain any connection to proposal, to applicant, co-applicant or subcontractor or to submitting institution (write "none" if no connection):

none

Review:

Please provide an overall evaluation summary rating:

<u>Excellent:</u> outstanding in all respects;
<u>Good:</u> quality but some deficiencies;
<u>Poor:</u> serious deficiencies.

Overall Evaluation Summary Rating	**Provide a brief explanation of your summary rating**
-Excellent	**This is a very strong proposal from a group of highly respected investigators andstrong institutional support. This proposal has a high likelihood for success, responds to the needs of CalFed, and is well justified fiscally. The suite of biomarkers and the linkages of responses to residue data in caged fish is of high merit. Some concerns with the potentials for mortality in the field, and also interpretation of the experimental biomarker data are noted.**
XGood	
-Poor	

1. **<u>Goals.</u>** Are the goals, objectives and hypotheses clearly stated and internally consistent? Is the concept timely and important?

   The goals, and objectives of this proposal are clearly stated and are internally consistent. Although the proposal is not hypothesis-based, the implied hypothesis is that caged fish in the impacted areas will show strong positive correlations among contaminant levels and positive responses in the boiomarker assays. Specifically, these linkages include lipophilic organic chemicals and CYP 1A induction, hormonally active compounds and CYP19 induction, trace metals and metallothionein induction, and overall contaminant levels and DNA and stress protein respones. The concept is certainly timely.

2. **Justification.** Is the study justified relative to existing knowledge? Is a conceptual model clearly stated in the proposal and does it explain the underlying basis for the proposed work? Is the selection of research, pilot or demonstration project, or a full-scale implementation project justified?

   Overall, this is a well justified proposal that is based upon the most current scientific techniques and knowledge. The conceptual model is clearly stated in the proposal. The proposed research is more of a phase I or pilot project in nature, as additional follow-up studies work will be necessary to help fine-tune the biomarker responses and to directly related biochemical and molecular changes to higher level changes that directly relate to the fitness of the salmon.

3. **Approach.** Is the approach well designed and appropriate for meeting the objectives of the project? Are results likely to add to the base of knowledge? Is the project likely to generate novel information, methodology or approaches? Will the information ultimately be useful to decision-makers?

   This is a well designed study and appropriate for meeting the objectives of the project. Because this project employs standard biochemical and molecular assay methodologies, this project is not likely to generate novel approaches. However, the information gained on using in situ caged fish, residue analysis, and a combination of molecular and biochemical markers should generate highly useful scientific information and publications. It should be noted that some of the biomarker responses can be affected by stress, oxygen levels, the presence of natural products, and toxicant interactions. Also, as mentioned, the current study does not employ any attempt to link the biomarker respone data to fish health or fitness parameters. Accordingly, the information gathered may be of limited initial use to decision makers. This does not detract for enthusiasm for the proposal, however, as it is very important to initially establish some cause and effect relationships and potential contaminants of biological concern prior to additional study.

   The combination of biomarker approaches used, DNA strand breaks, DNA content variation, acetylcholinsterase, stress proteins, and CYP responses are well justified and reflect biomarkers of both exposure and effects. One question is with regards to modulation of some of the CYP450 isozymes. For example, what is known about the induceability of CYP4B/F in the liver and CYP19 induction in the ovary and testis of salmon smolts? Has it been demonstrated that these two isozymes are readily induced in salmon exposed to the type of contaminants present in the sampling areas? This is a very important question, as the authors do not present preliminary data here. If these enzymes are not highly inducible, then it likely that no effects on these parameters will be observed, and such data could confound interpretation of the data.

   With regards to metallothionein induction, it should be indicated as to which salmon metallothionein (if more than one present) are going to be measured and which MT antibody used. Tis may be a dumb question as the reviewer is not very knowledgeable of salmon MTs. Vertebrates, in general, may express different metallothionein proteins which show varying degrees of induceability upon metal exposure. There is some other information that does not appear to be present in the proposal that should be clarified. For example are the ages of the hatchery reared salmon smolts to be placed at the three stations and in the San Joaquin River generally similar to those of wild fish that actually migrate through these sites at these times ? It is assumed that they are. This is important, as different developmental stages can effect susceptibility is to environmental chemicals.

4. **Feasibility.** Is the approach fully documented and technically feasible? What is the likelihood of success? Is the scale of the project consistent with the objectives?

This is a highly feasible project. However, as the authors clearly indicate that in a prior feasibility study of caging fish at these sites, a significant to amount of mortality occurred. The mortality was likely result of severe weather and temperature conditions during the previous study that are not anticipated to reoccur. Still, it is possible that some mortality will occur during the caging of fish will occur. it wasnt clear in the proposal if there were 3 cages of n=10 fish per site, or one cage. if one cage, is it feasible fto place an additional cage or more fish per site to allow for mortality ? this may not be feasible, but mortality could strongly influence statistical power and ability to discriminate biomarker changes. perhaps addition of a cage or additional fish at the high impact sites would be helpful.

5. **Project-Specific Performance Measures.** Does the project include appropriate performance measures to measure success relative to the project's goals and objectives? Is there enough detail as to how the performance measures will be quantified? For restoration projects, are monitoring plans explicit and detailed enough to determine if performance measures will be adequately assessed?

   There are appropriate performance measures indicated, and theseare relatively straightforward. However, it is not entirely clear as to 1) the extent of inhibition or induction each biomarker response are expected, or as the relevance of altered biomarker expression. For example would a 50% increase in CYP1A induction associated with organic chemical exposure be considered a significant biological response? Also, the presence of metals may confound some of the CYP responses in fish. Accordingly interpretation of some of the data may be difficult. These limitatiions are typical of field biomarker studies and do not necessarily reflect shortcomings in the experimental design. In addition, these limitations will be somewhat balanced by the incorporation of a suite of markers in the present study, some of which will likely be more robust than others. In particular indices on acetylcholinesterse inhibition and DNA injury will directly reflect the adverse effects of contaminant exposure on these fish.

6. **Products.** Are products of value likely from the project? Specifically for restoration projects, are products of value also likely from the monitoring component? Are interpretative outcomes likely from the project?

   This research project will generate important data relative to the effects of contaminant exposure in salmon smolts. Interpretive outcomes will be generated.

7. **Capabilities.** What is the track record of applicants in terms of past projects? Is the project team qualified to efficiently and effectively implement the proposed project? Do they have available the infrastructure and other aspects of support necessary to accomplish the project?

   The investigators are well respected in the academic communty and have considerable expertise in the techniques involved. The laboratories appear to be an well-equipped and the resources at the University of California Davis and Bodega Bay are excellent

8. **Cost/Benefit Comments.** Is the budget reasonable and adequate for the work proposed?

   The budget appears to be in reasonable and well justified with regards to the scope of work. It is a bit unclear as to the nature of the enzyme activity/ histochemistry student remission for one-quarter @ $1,685 .

Miscellaneous comments:

External Scientific: #4

Research and Restoration External Scientific Review Form

Proposal Number: **214**

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Conflict of Interest Statements:
I have no financial interest in this proposal.
XCorrect
-Incorrect

In the blank below please explain any connection to proposal, to applicant, co-applicant or subcontractor or to submitting institution (write "none" if no connection):

None

Review:

Please provide an overall evaluation summary rating:

Excellent: outstanding in all respects;
Good: quality but some deficiencies;
Poor: serious deficiencies.

Overall Evaluation Summary Rating	**Provide a brief explanation of your summary rating**
-Excellent	**I highly recommend funding that part of the project in which tissue burdens of contaminants will be measured. However, until this data is at hand and shows differences among exposure groups, I cannot support funding of the biomarker studies. Also, if funded, it is recommended that the PI consider some revisions to the biomarker studies before their implementation .**
XGood	
-Poor	

1. **Goals.** Are the goals, objectives and hypotheses clearly stated and internally consistent? Is the concept timely and important?

   Yes, the goals, objectives, and hypotheses are clearly stated. An evaluation of the effects of contaminants on the health of fish populations in the Delta is timely, important, and consistent with the goals of CalFed's Ecosystem Restoration Program. I'm just not sure that this is a good model to address this issue.

2. **Justification.** Is the study justified relative to existing knowledge? Is a conceptual model clearly stated in the proposal and does it explain the underlying basis for the proposed work? Is the selection of research, pilot or demonstration project, or a full-scale implementation project justified?

That is not clear from the proposal. The study is based on the observation of reduced survivorship of chinook salmon smolts that use the Old River compared to those that use the main stem San Joaquin River during downriver migrations. However, the data is from a non-peer reviewed report supported by the EPA and was not made available to this reviewer. Also, the report published in 1992 was probably from data collected in the late 1980s or early 1990s. Thus, it is not clear if this phenomenon still occurs. I was very surprised that the PI did not provide more data on the extent of this difference in survivorship and its temporal stability given its importance in justifying this project. Thus, it is difficult for this reviewer to evaluate the use of this model to evaluate the effects of contaminants on fish populations in the Delta. Without this information, it is difficult to defend the support of a full-fledged research project on this question.

3. **Approach.** Is the approach well designed and appropriate for meeting the objectives of the project? Are results likely to add to the base of knowledge? Is the project likely to generate novel information, methodology or approaches? Will the information ultimately be useful to decision-makers?

   Yes and no. Certainly it is important and will be informative to quantify tissue burdens of contaminants in these fish from the two overall sites. This information will be very useful for decision-makers. However, the use of biomarker approaches may be premature. Apparently, the preliminary study executed this past year showed very little difference in tissue burdens of pesticides or metals between fish caged in the Old River and San Joaquin. If there are no significant differences in tissue burdens, why go on and do biomarker studies. Maybe instead levels of other contaminants should be investigated. What will positive biomarker responses mean in the absence of chemical data showing significant differences between the two rivers?

   The water and tissue burden data generated from this project will be new and informative.

   The project will not generate any new methodologies or approaches. All of the techniques to be applied have been in the arsenal of ecotoxicologists for a long time. This is a shotgun approach with the hope that something interesting will come out.

4. **Feasibility.** Is the approach fully documented and technically feasible? What is the likelihood of success? Is the scale of the project consistent with the objectives?

   Yes the approach is fully documented and is technically feasible. Having said that, this reviewer has several major problems with the project design.

   1. The number of fish to be caged at each site at each time (n=10) is woefully small. Interindividual variation in biomarker responses is usually very large. These sample sizes will make it very difficult to demonstrate statistical significance between exposure groups. Also, it is likely that some mortality will occur, further reducing sample sizes and accentuating the problem.

   2. Exposure times of four days may be too brief to evoke significant responses at some of the biomarker endpoints. Induction of some genes with some contaminants, depending upon tissue, takes weeks and even longer. It all depends on the toxicokinetics of the contaminants, which in this case are largely unknown. Also, because of the short exposure and thus acclimation times, stress from handling or caging may influence some of the biomarker responses.

3. Several of the biomarker responses (heat shock protein and metallothionein expression) are subject to a variety of stressors not associated with chemical exposures. These might include handling of fish, temperature, caging, O2 stress, etc. Thus, it will be very difficult to determine if the response is due to chemical exposure or other environmental factors. Of course, this results in high levels of interindividual variation in responses at these endpoints. In fact, the PI indicates that levels of DNA damage (which you wouldn't expect to vary) in salmon smolts were elevated by the handling of the fish in preliminary experiments.

4. Few of these proposed biomarkers are mechanistically linked. I expect that some biomarker responses will be up and others down in fish caged in the Old River. How will these discordant data be interpreted? Although the Data analysis section mentions that ANOVA will be used to evaluate for differences at each biomarker endpoint, how will the entire data set be investigated for trends, patterns, etc. Little attention is paid to putting the whole package together. In the end, it will be difficult to determine whether there is a problem or not in the Old River.

5. What are the control samples? It is mentioned that biomarker responses will be compared to controls. It is possible that responses in the San Joaquin at some endpoints may be higher than in the Old River. Better controls are needed.

6. Cytochrome P4501A expression is well studied in the fish literature and is known to be significantly inducible by a variet y of aromatic hydrocarbon contaminants. However, I'm not sure that the other two CYPs to be measured are highly inducible in fish. No references were provided that report their inducibility in fish. Even if they are in mammals (and I'm not sure that they are), that doesn't mean that they are inducible in fish. For example, CYP1B1 is highly inducible in mammals by TCDD, but not in the two species of fish investigated. Additionally, why are the investigators developing an antibody probe for CYP1A. The available probes from scup and Atlantic cod both work real well on almost all fish taxa.

7. Metallothionein is the exact opposite story from CYP1A1. Do the investigators have a metallothionein probe and are the sure that it will work with chinook salmon? Metallothionein is quite tricky with which to work and interpret the results. Many studies in fish have questioned the use of MT as an effective biomarker in the absence of thorough controlled laboratory studies. Also, there is no mention in the Methods as to which MT probe will be used.

5. **Project-Specific Performance Measures.** Does the project include appropriate performance measures to measure success relative to the project's goals and objectives? Is there enough detail as to how the performance measures will be quantified? For restoration projects, are monitoring plans explicit and detailed enough to determine if performance measures will be adequately assessed?

   The project was sufficiently structured such that it will be easy to ascertain if its goals and objectives are obtained. I'm certain that each of objectives will be completed in a timely fashion.

6. **Products.** Are products of value likely from the project? Specifically for restoration projects, are products of value also likely from the monitoring component? Are interpretative outcomes likely from the project?

   The chemical data will be valuable, but I doubt that most of the biomarker responses will prove informative. As I mentioned earlier, it will be very difficult to interpret the meaning of the biomarker responses.

7. **Capabilities.** What is the track record of applicants in terms of past projects? Is the project team qualified to efficiently and effectively implement the proposed project? Do they have available the infrastructure and other aspects of support necessary to accomplish the project?

   Yes, the applicants should be able to successfully complete this project as outlined based on their past experiences and infrastructure.

8. **Cost/Benefit Comments.** Is the budget reasonable and adequate for the work proposed?

   The budget is reasonable given the number of diverse tasks to be performed, but given the small sample numbers (n=180 in total), I'm not sure that this expenditure is justified. As a small aside, I don't understand why 20 boat days (at $384/day) are required to deploy and retrieve cages at two on three occasions.

Miscellaneous comments:

As an aside, I would recommend that telemetry studies be conducted on smolts from the natural population of salmon, to compare their residence times and behaviors in the Old River and San Joaquin River.

Environmental Compliance:

Proposal Number: 214

Applicant Organization: University of California, Davis, Bodega Marine Laboratory

Proposal Title: Decreased Survival of Chinook Salmon Smolt in the Old River: Biological Responses to Contaminants

1. Are the legal or regulatory issues that affect the proposal identified adequately in the proposal?

   XYes **-**No

   If no, please explain:

2. Does the project's timeline and budget reflect adequate planning to address legal and regulatory issues that affect the proposal?

   XYes **-**No

   If no, please explain:

3. Do the legal and regulatory issues that affect the proposal significantly impair the project's feasibility?

   -Yes **X**No

   If yes, please explain:

Other Comments:

Budget:

Proposal Number: 214

Applicant Organization: University of California, Davis, Bodega Marine Laboratory

Proposal Title: Decreased Survival of Chinook Salmon Smolt in the Old River: Biological Responses to Contaminants

1. Does the proposal include a detailed budget for each year of requested support?

   XYes **-**No

   If no, please explain:

2. Does the proposal include a detailed budget for each task identified?

   XYes **-**No

   If no, please explain:

3. Does the proposal clearly state the type of expenses encompassed in indirect rates or overhead costs?

   -Yes **X**No

   If no, please explain:

   UC Davis overhead rate 10% not 26% on State Contracts.

4. Are appropriate project management costs clearly identified?

   XYes **-**No

   If no, please explain:

5. Do the total funds requested (Form I, Question 17A) equal the combined total annual costs in the budget summary?

   XYes **-**No

   If no, please explain (for example, are costs to be reimbursed by cost share funds included in the budget summary).

6. Does the budget justification adequately explain major expenses?

   XYes **-**No

If no, please explain:

7. Are there other budget issues that warrant consideration?

   -Yes **X**No

   If yes, please explain:

Other Comments: